Impact Objectives

- Produce and test a new microsystem technology for drug screening of human intracellular ion channels
- Investigate two proteins that are involved in cancer and neurodegeneration (CLICL1 and CLIC4)

Innovations in drug screening

By bridging the fields of microfluidic engineering and biochemistry, ***Dr Michele Zagnoni*** *and* ***Dr Cheryl Woolhead*** *have developed a novel and efficient method to discover and test new medications*

Dr Michele Zagnoni

Dr Cheryl Woolhead

Could you introduce the type of research conducted by your respective laboratories?

MZ: The Zagnoni Lab is based at the University of Strathclyde in Glasgow in the UK, in the Electronic and Electrical Engineering Department. Our expertise is in the development of microfluidic technologies for healthcare applications, including fundamental biological research, drug screening, personalised medicine therapy, organ-on-a-chip and artificial cell membrane constructs. As such, we bring to this project multidisciplinary expertise to create biosensors and screening platforms that are suitable for academic and industrial research.

CW: The Woolhead Lab is based at the University of Glasgow in the Institute of Molecular, Cell and Systems Biology. Our expertise is in the analysis of membrane proteins from synthesis to integration into the target lipid bilayer. As such, we bring to this project an understanding of the molecular properties of the proteins we are studying and the physiological conditions to optimise protein folding and integration.

You are currently developing a droplet-based microfluidic platform for intracellular ion channel drug discovery. How did you come to establish this project?

MZ: The Zagnoni Lab have long been working on microfluidic artificial cell membranes and we were interested in synthesising proteins within a droplet-based system for facilitating drug screening studies. The Woolhead group has for many years been working on the development of a cell-free system for protein synthesis, to allow incorporation of modified aminoacyl tRNA molecules. Utilising a cell-free system such as this for high-throughput screening will increase the speed and lower the cost of such testing, allowing many more targets to be screened against a drug library. To achieve this goal an interdisciplinary approach was needed, which brought our two groups together to collaborate.

Why did you decide to focus on two proteins – CLIC1 and CLIC4 – specifically?

CW: The CLIC proteins are of great interest to us due to their reported involvement in various disease states, from neurodegenerative conditions to cancer. Whereas a potential inhibitor has been reported for CLIC1, no such inhibitors have been identified for CLIC4, therefore for us the challenge is to dissect our knowledge of CLIC1 and implement it into the study of CLIC4, underpinning the basis for new drug discovery approaches in the future for intracellular ion channels.

The project is approximately midway through its three-year funding period. What progress has been made so far?

MZ: We have made great progress both in developing a robust microfluidic system for testing a variety of ion channels and in the preparation and purification of the CLIC proteins. Our preliminary results have shown our prototype is working and our assays have produced interesting mechanistic results. These will now be expanded on through drug testing in conjunction with our industrial partners.

Do you have any plans for future projects that you would like to share with readers?

MZ: Building on the outcomes from this project, we will implement the use of cell-free expression in microfluidic systems to create artificial cells and explore several cell membrane associated processes, advancing our understanding of how membrane proteins function. In particular, we aim to address how viral ion channels function and translate the current technology for the study of more complex membrane proteins, such as GPCR channels.

A faster way to new medications

*Researchers at the **University of Strathclyde** and **University of Glasgow** in the UK, have joined forces to develop an innovative method to test new drugs prior to clinical trials using synthetic biology. Their research has major implications for pharmaceutical drug discovery of medications targeting ion channels*

Ion channels are membrane proteins, which generate pores that ions can pass through. Ion channels play an important role within our bodies, as they determine the physical processes that our tissues go through – from whether we will develop a pathological condition to simply whether our heart beats or nervous system operates. Since ion channels are frequently targeted by pathogens and other substances, they are of major research interest to those involved in developing pharmaceuticals.

Drug discovery is the process by which new candidate medications are discovered. Historically, screening for drugs targeting ion channels has been a slow and expensive process. Now, however, a collaborative research project involving the University of Strathclyde and the University of Glasgow could provide an alternative to such methods, pushing the pharmaceutical market in a novel and more effective direction.

A DIFFERENT SYSTEM

Ion channels are the second largest class of pharmacological drug targets, meaning they are key players in developing new medications. In conventional drug discovery, an automated patch-clamp method is used to study the electrophysiological currents through ion channels and therefore identify drugs of interest. However, this method cannot be used to study intracellular ion channels, meaning a whole slew of therapeutic potential cannot be adequately investigated.

Dr Michele Zagnoni of the University of Strathclyde and Dr Cheryl Woolhead of the University of Glasgow have come together to produce and test a new microsystem technology aimed at screening drugs using intracellular ion channels. They are working to develop microfluidic artificial cell membranes and a cell-free system of protein synthesis. Their goal is to increase the speed and lower the cost of drug screening. 'Microfluidics offers the ability to create synthetic cells that can be used for drug screening, avoiding higher costs and challenges implicated with live cell measurements,' elaborates Zagnoni.

By means of these microfluidic technologies and cell-free protein synthesis, their research offers a controlled way to create *in vitro* models of biological systems. By using these models, they can use intracellular ion channels to identify, discover and develop new chemical compounds for medications. Because these procedures take place before clinical trials, their increased efficiency identifying drugs is expected to improve outcomes of the clinical trials that follow.

INNOVATIVE COLLABORATION

The Zagnoni Lab is focused on bioengineering, while the Woolhead Lab is focused in molecular biochemistry. 'The work carried out in the two laboratories is very different,' explains Woolhead. 'As such, this partnership allows us to think about the problem much more laterally, from analysing the most efficient way to create our tests, to the most physiological way and finding a constructive path through the middle.'

Both laboratories are located in Glasgow and their proximity to one another has enabled the labs to develop a close relationship alongside a state-of-the-art approach to drug discovery. The Zagnoni Lab focuses on microfluidic technology, screening compounds on human tissue and assessing characteristics of cellular function in microfluidic environments. At the same time, the Woolhead Lab performs in-depth analysis of membrane proteins, from their synthesis to their integration into the lipid bilayer. Woolhead has also published research on the early stages of protein folding and is currently studying structural changes in proteins during lipid integration.

The novelty of Zagnoni and Woolhead's work is certainly in their collaboration. By combining biochemistry and microfluidic engineering, they have been able to make novel advances in synthetic biology.

MOVING FORWARD

So far, Zagnoni and Woolhead are on track to reach their goal of establishing a microfluidic platform for intracellular ion channel drug discovery. The next step is to implement their microfluidic system into a semi-automatic prototype suitable for drug screening using artificial cell membranes.

At present, they are investigating CLIC1 and CLIC4, two proteins implicated in a range of pathological conditions. While there is a known inhibitor for CLIC1, this is not the case for CLIC4. Future research will aim to identify a compound that would modulate the activity of CLIC4 ion channels. Such a discovery would be the first of its kind. Additionally, Zagnoni and Woolhead are working with two industrial partners, ▶

Microfluidics offers the ability to create synthetic cells that can be used for drug screening, avoiding higher costs and challenges implicated with live cell measurements

Apconix and Smartox. Their partnership with pharmaceutical companies will allow them to test their method using current pharmaceutical drug screening scenarios, as well as test their system with animal-derived molecules, which could lead to novel drug identification.

MULTI-LAYERED EFFECTS

Zagnoni and Woolhead's research is expected to have positive consequences across not only industrial, but also academic, economic and societal realms. From an academic standpoint, their research is seeking to enhance understanding of membrane proteins, specifically the mechanisms of intracellular membrane protein structures. They are also looking into the functional role of controllable artificial cell membranes. These studies could open up a plethora of research opportunities. Zagnoni and Woolhead are already working hard to disseminate their project outputs and develop relationships with labs around Europe, and so maximise their research impact.

Economically, this new system of drug screening would allow a decrease in developmental expenditure of certain pharmaceuticals. This would make clinical trials more affordable, allowing new and necessary medications to be available more quickly. The preclinical phase, where new compounds are discovered and identified, accounts for a third of the total cost of clinical trials. By making savings at this stage, total clinical trial expenditure is likely to be significantly reduced.

Most importantly, by developing an inventive and efficient way to perform drug testing, there will be considerable societal implications. Because these initial screenings will lead to new economic and effective medications, patients should have greater access to vital interventions. This means reduced mortality and morbidity and improved quality of life for patients.

By merging fields, Zagnoni and Woolhead have made leaps and bounds toward developing a microfluidic platform for intracellular ion channel drug discovery. Their innovative use of synthetic biology is likely to facilitate drug testing at a higher throughput – paving the way for more thorough, fast and effective processing of new medications and pharmaceutical innovations than ever before. ●

Project Insights

FUNDING

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BIOGRAPHIES

Dr Michele Zagnoni is a Senior Lecturer and Group Leader of the Microfluidic Group in the Electronic and Electrical Engineering Department at the University of Strathclyde, Glasgow. He has extensive expertise in bridging microfluidic technology to biological applications in the areas of cancer research, neuroscience and ion channel characterisation. Recently, he became the Chief Scientific Officer of ScreenIn3D, a joint venture company that provides drug screening services based on microfluidics and 3D cell culture.

Dr Cheryl Woolhead is the Deputy Director of the Institute of Molecular, Cell and Systems Biology in the College of Medical, Veterinary and Life Sciences at the University of Glasgow. She has worked on membrane proteins and their structures for over 20 years, focusing on the early folding of proteins within the ribosome tunnel and investigating how ribosome/nascent chain interactions affect translation and cellular targeting.